

IN THE CLAIMS:

1. (CURRENTLY AMENDED) A bioreactor for producing functional cartilaginous tissue from a cell-seeded scaffold ~~or a cell-seeded scaffold integrated with an osteoconductive and/or osteoinductive substrate~~, comprising:

(a) a growth chamber; and

(b) means for applying strain-controlled deformational loading ~~via loading platens to the cell-seeded scaffold or cell-seeded scaffold integrated with an osteoconductive and/or osteoinductive substrate~~; and

~~wherein~~ (c) means for controlling the strain-controlled deformational loading ~~is controlled~~ according to a loading regime for ~~facilitating~~ producing functional cartilaginous tissue ~~growth~~.

2. (ORIGINAL) The bioreactor of Claim 1, wherein the scaffold is bioresorbable.

3. (ORIGINAL) The bioreactor of Claim 1, wherein the scaffold is biocompatible.

4. (ORIGINAL) The bioreactor of Claim 1, wherein the scaffold is biodegradable.

5. (ORIGINAL) The bioreactor of Claim 1, wherein the scaffold is non-bioresorbable.

6. (CURRENTLY AMENDED) The bioreactor of Claim 62, wherein means ~~(b) further~~ (d) applies intermittent cyclical hydrostatic fluid pressure to the cell-seeded scaffold.

7. (CURRENTLY AMENDED) The bioreactor of Claim 6, wherein ~~the~~ means (d) is capable of applying hydrostatic fluid is ~~pressurized~~ pressure at from about 0 to about 18 MPa.

8. (CURRENTLY AMENDED) The bioreactor of Claim 7, wherein the hydrostatic fluid ~~is pressurized at~~ is applied at a pressure of from about 0 to about 6 MPa.

9. (CURRENTLY AMENDED) The bioreactor of Claim 6, wherein ~~the~~ means (d) is capable of applying hydrostatic fluid pressure at a cyclical frequency is of from about 0 to about 5 Hz.

10. (CURRENTLY AMENDED) The bioreactor of Claim 9, wherein ~~the~~ hydrostatic fluid pressure is applied at a cyclical frequency is of from about 0.1 to about 2 Hz.

11. (CURRENTLY AMENDED) The bioreactor of Claim 6, wherein ~~the~~ means (d) is capable of applying hydrostatic fluid pressure is ~~applied~~ for from about 0.5 to about 18 hours per day.

12. (CURRENTLY AMENDED) The bioreactor of Claim 11, wherein the hydrostatic fluid pressure is applied for from about 2 to about 6 hours per day.

13. (CURRENTLY AMENDED) The bioreactor of Claim 1, wherein means (b) ~~applies~~ is capable of applying intermittent cyclical strain-controlled deformational loading.

14. (CURRENTLY AMENDED) The bioreactor of Claim 13, wherein the strain-controlled deformational loading ~~is~~ can be applied at from about 0 to about 50% strain, based upon the thickness of the cell-seeded scaffold.

15. (CURRENTLY AMENDED) The bioreactor of Claim 14, wherein the strain-controlled deformational loading is can be applied at from about 0 to about 20% strain, based upon the thickness of the cell-seeded scaffold.

16. (CURRENTLY AMENDED) The bioreactor of Claim 13, wherein the ~~eyclical frequency of~~ strain-controlled deformational loading is can be applied at a cyclical frequency of from about 0 to about 5 Hz.

17. (ORIGINAL) The bioreactor of Claim 16, wherein the strain-controlled deformational loading can be applied at a cyclical frequency is of from about 0.1 to about 2 Hz.

18. (CURRENTLY AMENDED) The bioreactor of Claim 13, wherein the strain-controlled deformational loading is can be applied for from about 0.5 to about 18 hours per day.

19. (CURRENTLY AMENDED) The bioreactor of Claim 18, wherein the strain-controlled deformational loading is can be applied for from about 2 to about 6 hours per day.

20. (CURRENTLY AMENDED) The bioreactor of Claim 1, wherein means (b) applies is capable of applying intermittent cyclical hydrostatic fluid pressure and which also comprises means (d) for applying intermittent ~~strain-controlled~~ strain-controlled cyclical deformational loading.

21. (PREVIOUSLY PRESENTED) The bioreactor of Claim 20, wherein the amplitude of the cyclical hydrostatic fluid pressure and the amplitude of the strain-controlled deformational loading are modified over time as matrix elaboration proceeds.

22. (CURRENTLY AMENDED) The bioreactor of Claim 1, wherein the ~~resulting~~ functional cartilaginous tissue comprises hyaline cartilage.

23. (CURRENTLY AMENDED) The bioreactor of Claim 1, wherein the ~~resulting~~ functional cartilaginous tissue comprises hyaline cartilage and an osteoconductive and/or osteoinductive substrate.

24. (CURRENTLY AMENDED) The bioreactor of Claim 1, wherein the ~~resulting~~ functional cartilaginous tissue comprises elastic cartilage.

25. (CURRENTLY AMENDED) The bioreactor of Claim 1, wherein the ~~resulting~~ functional cartilaginous tissue comprises fibrocartilage.

26. (CURRENTLY AMENDED) The bioreactor of Claim 1; which also comprises means for producing tissue in a desired ~~shapes~~ shape.

27. (CURRENTLY AMENDED) The bioreactor of Claim 26, wherein the ~~shaped tissue~~ desired shape conforms to a body part, a prosthesis, a cosmetic implant, or a defect to be filled.

28. (CURRENTLY AMENDED) The bioreactor of Claim 1, wherein ~~the~~ means (b) comprises loading platens which ~~produce deformational loading~~ conform to a body part, a prosthesis, a cosmetic implant, or a defect to be filled.

29. (CURRENTLY AMENDED) A method for producing functional cartilaginous tissue from a cell-seeded scaffold, ~~or a cell-seeded scaffold integrated with an osteoconductive and/or osteoinductive substrate, said~~ the method comprising the steps of:

(a) ~~inoculating chondrocytes or chondroprogenitors into a scaffold or a scaffold integrated with an osteoconductive and/or osteoinductive substrates to form a~~ cell-seeded scaffold;

(b) placing the cell-seeded scaffold ~~or cell-seeded scaffold integrated with an osteoconductive and/or osteoinductive substrate~~ into a bioreactor;

(c) filling said bioreactor with a liquid growth medium;

(d) applying strain-controlled deformational loading to the cell-seeded scaffold ~~or cell-seeded scaffold integrated with an osteoconductive and/or osteoinductive substrate via a loading platens~~ according to a loading regime ~~optimized for~~ producing functional cartilaginous tissue growth; and

(e) culturing said ~~cell-seed scaffold or cell-seeded scaffold integrated with an osteoconductive and/or osteoinductive substrate from step (d) for a time sufficient to produce~~ for producing functional cartilaginous tissue.

30. (CURRENTLY AMENDED) The method of Claim 29, wherein the bioreactor comprises:

(a) a growth chamber; and

(b) means for applying strain-controlled deformational loading ~~via loading platens~~ to the cell-seeded scaffold ~~or cell-seeded scaffold integrated with an osteoconductive and/or osteoinductive substrate;~~ and

~~wherein~~ (c) means for controlling the strain-controlled deformational loading ~~is controlled~~ according to a loading regime for ~~facilitating~~ producing functional cartilaginous tissue growth.

31. (ORIGINAL) The method of Claim 29, wherein the scaffold is biocompatible.

32. (ORIGINAL) The method of Claim 29, wherein the scaffold is biodegradable.

33. (ORIGINAL) The method of Claim 29, wherein the scaffold is non-biodegradable.

34. (ORIGINAL) The method of Claim 29, wherein the scaffold is bioresorbable.

35. (ORIGINAL) The method of Claim 29, wherein said stressed cells:
(a) display enhanced maintenance of a chondrocyte phenotype; and
(b) produce a functional cartilaginous matrix.

36. (CURRENTLY AMENDED) The ~~method~~ bioreactor of Claim 29 ~~62~~, wherein ~~in step (d) hydrostatic fluid pressure is further applied by means comprising (d)~~ comprises a reservoir, a pump, and tubing interconnecting said growth chamber, said reservoir, and said pump, so as to allow pressurization of liquid growth medium from said reservoir, in response to force applied by said pump.

37. (CURRENTLY AMENDED) The ~~method~~ bioreactor of Claim 36, wherein said pump comprises a piston and chamber.

38. (CURRENTLY AMENDED) The method of Claim 63, wherein ~~in step (d)~~ intermittent cyclical hydrostatic fluid pressure is also applied to the cell-seeded scaffold.

39. (CURRENTLY AMENDED) The method of Claim 38, wherein the hydrostatic fluid is ~~pressurized at~~ applied at a pressure of from about 0 to about 18 MPa.

40. (CURRENTLY AMENDED) The method of Claim 39, wherein the hydrostatic fluid is ~~pressurized at~~ applied at a pressure of from about 0 to about 6 MPa.

41. (CURRENTLY AMENDED) The method of Claim 38, wherein the ~~eyelical frequency~~ hydrostatic fluid pressure is applied at a cyclical frequency of from about 0 to about 5 Hz.

42. (CURRENTLY AMENDED) The method of Claim 41, wherein the ~~eyelical frequency~~ hydrostatic fluid pressure is applied at a cyclical frequency of from about 0.1 to about 2 Hz.

43. (CURRENTLY AMENDED) The method of Claim 29 ~~38~~, wherein the hydrostatic fluid pressure is applied for from about 0.5 to about 18 hours per day.

44. (PREVIOUSLY PRESENTED) The method of Claim 43, wherein the fluid pressure is applied for from about 2 to about 6 hours per day.

45. (CURRENTLY AMENDED) The method of Claim 29, wherein ~~in step (d)~~ intermittent cyclical strain-controlled deformational loading is applied.

46. (PREVIOUSLY PRESENTED) The method of Claim 45, wherein the strain-controlled deformational loading is from about 0 to about 50% strain, based upon the thickness of the cell-seeded scaffold.

47. (CURRENTLY AMENDED) The method of Claim 46, wherein the strain-controlled deformational loading is from about 0 to about 20% strain, based upon the thickness of the cell-seeded scaffold.

48. (CURRENTLY AMENDED) The method of Claim 45, wherein the ~~eyelical frequency of~~ strain-controlled deformation loading is applied at a cyclical frequency of from about 0 to about 5 Hz.

49. (CURRENTLY AMENDED) The method of Claim 48, wherein the strain-controlled deformational loading is applied at a cyclical frequency is of from about 0.1 to about 2 hz.

50. (CURRENTLY AMENDED) The method of Claim 45, wherein the strain-controlled deformational loading is applied for from about 0.5 to about 18 hours per day.

51. (CURRENTLY AMENDED) The method of Claim 50, wherein the strain-controlled deformational loading is applied for from about 2 to about 6 hours per day.

52. (CURRENTLY AMENDED) The method of Claim 29, wherein ~~in step (d)~~ intermittent cyclical hydrostatic fluid pressure and intermittent cyclical strain-controlled deformational loading are applied to the cell-seeded scaffold.

53. (PREVIOUSLY PRESENTED) The method of Claim 52, wherein the amplitude of the cyclical hydrostatic pressure and the amplitude of the deformational strain-controlled loading are modified over time as matrix elaboration proceeds.

54. (CURRENTLY AMENDED) The method of Claim 29, wherein the ~~resulting~~ functional cartilaginous tissue comprises hyaline cartilage.

55. (CURRENTLY AMENDED) The method of Claim 29, wherein the ~~resulting~~ functional cartilaginous tissue comprises hylaine cartilage and osteoinductive substrate.

56. (CURRENTLY AMENDED) The method of Claim 29, wherein the ~~resulting~~ functional cartilaginous tissue comprises elastic cartilage.

57. (CURRENTLY AMENDED) The method of Claim 29, wherein the ~~resulting~~ functional cartilaginous tissue comprises fibrocartilage.

58. (CURRENTLY AMENDED) The method of Claim 29, wherein the bioreactor comprises means for producing tissue in a desired shape.

59. (CURRENTLY AMENDED) The bioreactor of Claim 29, where the strain-controlled deformational loading is applied via loading platens which ~~produce deformational loading~~ conform to a body part, a prosthesis, a cosmetic implant, or a defect to be filled.

60. (CURRENTLY AMENDED) The method of Claim ~~59~~ 58, wherein the ~~shaped tissue~~ desired shape conforms to a body part, a prosthesis, a cosmetic implant, or a defect to be filled.

61. (CURRENTLY AMENDED) A bioreactor system for producing functional cartilaginous tissue, comprising:

(a) a cell-seeded scaffold ~~or a cell-seeded scaffold integrated with an osteoconductive and/or osteoinductive substrate;~~

(b) a growth chamber; and

(c) means for applying strain-controlled loading to a cell-seeded scaffold ~~or a cell-seeded substance integrated with an osteoconductive and/or osteoinductive substance in the growth chamber via a loading platens;~~ and

wherein (d) means for controlling the strain-controlled deformational loading ~~are controlled~~ according to a loading regime for facilitating producing functional cartilaginous tissue ~~growth~~.

62. (CURRENTLY AMENDED) The bioreactor of Claim 1, ~~wherein which~~ also comprises means ~~(b) further applies (d) for applying~~ hydrostatic fluid pressure to the cell-seeded scaffold.

63. (CURRENTLY AMENDED) The method of Claim 29, ~~wherein in step (d)~~
which also comprises applying hydrostatic fluid pressure is also applied to the cell-seeded
scaffold.

64. (NEW) The bioreactor system of Claim 61 which also comprises means (e)
for applying hydrostatic fluid pressure to the cell-seeded scaffold.

65. (NEW) The bioreactor of Claim 1, wherein the cell-seeded scaffold is
integrated with an osteoconductive and/or osteoinductive substrate.

66. (NEW) The method of Claim 29, wherein the cell-seeded scaffold is
integrated with an osteoconductive and/or osteoinductive substrate.

67. (NEW) The bioreactor system of Claim 61, wherein the cell-seeded
scaffold is integrated with an osteoconductive and/or osteoinductive substrate.

68. (NEW) The bioreactor of Claim 1, wherein the strain-controlled
deformational loading is applied for up to 6 weeks.

69. (NEW) The bioreactor of Claim 62, wherein the hydrostatic fluid pressure
is applied for up to 6 weeks.

70. (NEW) The method of Claim 29, wherein the strain-controlled
deformational loading is applied for up to 6 weeks.

71. (NEW) The method of Claim 63, wherein the hydrostatic fluid pressure is
applied for up to 6 weeks.

72. (NEW) The bioreactor system of Claim 61, wherein the strain-controlled
deformational loading is applied for up to 6 weeks.

73. (NEW) The bioreactor system of Claim 64, wherein the hydrostatic fluid pressure is applied for up to 6 weeks.

74. (NEW) A bioreactor for producing functional cartilaginous tissue from a cell-seeded scaffold, comprising:

- (a) a growth chamber;
- (b) means for applying strain-controlled deformational loading to the cell-seeded scaffold;
- (c) means for applying intermittent cyclical hydrostatic fluid pressure to the cell-seeded scaffold; and
- (d) means for controlling the deformational loading and the hydrostatic fluid pressure according to a loading regime for producing tissue having the functional properties of cartilaginous tissue.

75. (NEW) A method for producing functional cartilaginous tissue from a cell-seeded scaffold, which method comprises:

- (a) inoculating chondrocytes or chondroprogenitors into a cell-seeded scaffold to form a cell-seeded scaffold;
- (b) placing the cell-seeded scaffold into a bioreactor;
- (c) filling said bioreactor with a liquid growth medium;
- (d) applying strain-controlled deformational loading to the cell-seeded scaffold according to a loading regime for producing functional cartilaginous tissue;
- (e) applying intermittent cyclical hydrostatic fluid pressure to the cell-seeded scaffold; and
- (f) culturing the cell-seed scaffold for a time sufficient to produce tissue having the functional properties of cartilaginous tissue.

76. (NEW) A bioreactor system for producing functional cartilaginous tissue, comprising:

- (a) a cell-seeded scaffold ;
- (b) a growth chamber;
- (c) means for applying strain-controlled loading to the cell-seeded scaffold in the growth chamber;
- (d) means for applying intermittent cyclical hydrostatic fluid pressure to the cell-seeded scaffold; and
- (e) means for controlling the deformational loading and the hydrostatic fluid pressure according to a loading regime for producing tissue having the functional properties of cartilaginous tissue.

77. (NEW) The bioreactor of Claim 1, wherein the strain-controlled deformational loading is applied at from about 0 to about 50% strain, based upon the thickness of the cell-seeded scaffold, at a cyclical frequency of from about 0 to about 5 Hz, for from about 0.5 to about 18 hours per day for up to 6 weeks.

78. (NEW) The bioreactor of Claim 62, wherein the hydrostatic fluid pressure is applied at from about 0 to about 18 MPa, at a cyclical frequency of from about 0 to about 5 Hz, for from about 0.5 to about 18 hours per day for up to 6 weeks.

79. (NEW) The method of Claim 29, wherein the strain-controlled deformational loading is applied at from about 0 to about 50% strain, based upon the thickness of the cell-seeded scaffold, at a cyclical frequency of from about 0 to about 5 Hz, for from about 0.5 to about 18 hours per day for up to 6 weeks.

80. (NEW) The method of Claim 63, wherein the hydrostatic fluid pressure is applied at from about 0 to about 18 MPa, at a cyclical frequency of from about 0 to about 5 Hz, for from about 0.5 to about 18 hours per day for up to 6 weeks.

81. (NEW) The bioreactor system of Claim 61, wherein the strain-controlled deformational loading is applied at from about 0 to about 50% strain, based upon the thickness of the cell-seeded scaffold, at a cyclical frequency of from about 0 to about 5 Hz, for from about 0.5 to about 18 hours per day for up to 6 weeks.

82. (NEW) The bioreactor system of Claim 64, wherein the hydrostatic fluid pressure is applied at from about 0 to about 18 MPa, at a cyclical frequency of from about 0 to about 5 Hz, for from about 0.5 to about 18 hours per day for up to 6 weeks.